REMARKS/ARGUMENTS

The Official Action has been carefully considered, and the Examiner's comments are duly noted. Reconsideration of this Application in view of the submission of new claims 24 to 35, cancellation of claims 1, 3-4, 6-14, 16, 22 and 23. The claims now in this Application are 24 to 35

The applicant acknowledges the withdrawal of method claims 1, 2, 5, 15 and 17-21 from prosecution herein.

The applicant hereby acknowledges the examination of the elected species of "dermal fibroblasts" or "fibroblasts".

With respect to the objection to the claims as indefinite, it is submitted that new claims 24 to 35 are free of these objections.

New claim 24 is intended to replace claim 3, and new claims 25 to 35 are now submitted for examination in this Application.

The new set of claims 24-35 are presented in order to define the product more specifically so that it is distinct from the cited prior art.

Claims Rejection 35 USC 102

1. Claims 3, 4, 6-14, 22 and 23 are rejected under USC 102 as being anticipated by US 5,755,814. (Berg. et. al)

The examiner contends that the Berg et al. patent discloses dermal fibroblasts preparation grown in culture vessels in basic DMEM with serum and in absence of matrices and that it anticipates the present invention in that the fibroblasts attached to plastic dishes proliferated better without matrices than the fibroblasts in presence of matrices.

Further the examiner also contends that the cited patent teaches that amount of cells in the final preparation depends on the cell density.

The applicant hereby wishes to present its clarifications.

The US Patent to Berg. et al. relates namely to a three dimensional system using a collagen sponge. More particularly referring to the example 3 of this cited patent, Berg. et al. have used cells grown on plastic dish without any matrix as a control to compare their three dimensional matrix. More precisely emphasizing, the cell grown on plastic without any matrix are cells which are routinely cultured as **monolayers** and they are usually taken as a control for comparisons.

The cells on plastic dishes are stated to be only proliferating better without a matrix. The cells on plastic dishes are **not disclosed nor anticipated to be** forming a three dimensional organization of cells. It is the cells on collagen sponge which is three dimensional in nature in the cited patent and not the cells on plastic dishes. Therefore, US patent 5755814 does not teach the formation of a three-dimensional organization without matrices, nor can this be anticipated from that invention.

Further in the cited patent of Berg .et al. the cells are seeded in a 24- well plate at $3x\ 10^5$ cells per well. As the diameter of a well in a 24-well plate is 1.5 cm, the area of each well is 1.76 cm^2. This gives a seeding density of 1.7×10^5 cells/ cm^2.

This cell density is much less than the lower limit of the macromass density range disclosed in the present application.

The applicant herein wishes to state that it cannot be anticipated from Berg et al. patent that higher densities will form three dimensional organization of cells. As the cited reference requires collagen matrix for forming three-dimensional construct, the present application does not require collagen matrix for three dimensional construct formation. The features of our application as compared to the cited reference of Berg. et al. is highlighted below:

Berg et. al	Present Interpretation	Distinct features of present invention
Cells on plastic dishes proliferate better	Not the main invention of the patent. The cells form a monolayer.	The present invention is three-dimensional organization of cells, without the aid of a matrix.
Amount of cells in the final preparation depends on cell density	The invention focuses on collagen matrix as inherent requirement, which then depends on the amount of cells.	The final preparation does not require collagen matrix for formation of three-dimensional construct.

2. Claims 3, 4, 6-14, 16, 22 and 23 are rejected under USC 102 as being anticipated by Furukawa et al.

The examiner contends that the Furukawa discloses three dimensional fibroblast aggregates obtained by seeding cells at "high density" and the final product of the Furukawa paper is similar to what is claimed in the present invention.

The examiner also states although the cited patent of Furukawa obtains the three dimensional aggregates by rotational culture the product is the same as the present application as the final components are not clearly defined by the present application.

The applicant herein has addressed the defining of the product of invention by amending the claims as required herein to overcome the previous objections. Now the final components of the claimed cellular product are defined in the claims.

In order to highlight the distinct features of the product of the present invention, the applicant herein describes the product and compares it with the product obtained by Furukawa et al.

The applicant herein states that the product of the present invention although named as a three dimensional organization of cells, is not similar to the three dimensional fibroblast aggregates of Furukawa et al. The product claimed in the present invention is

substantially different from and has unanticipated properties compared to the product in the Furukawa et al reference, as described in the following clarifications.

As it is evident from the figures or pictures of our construct, the present invention provides a product, which is a single, unified sheet like construct wherein the cells are uniformly distributed unlike the numerous aggregates, separated from each other, formed in the Furukawa et al paper.

Further the entire number of seeded cells in our invention become a single unified structure as against the cite Furukawa product wherein all the cells do not become a part of a single aggregate but they are distributed among numerous separated aggregates of cells.

The size of one aggregate in the cited reference of Furukawa is 200 to 280 um (0.2-0.28mm) which is an order of the magnitude smaller than the smallest macromass construct of 3-5 mm size and is two orders of magnitude smaller than 2.0 cm macromass construct shown in the figures. On the contrary, the present application provides construct, which is greater than the 3-5 mm in diameter and can be made as large as 2 cm in diam as shown in the figure.

The total number of cells in one aggregate is about 4000 cells (240 um) as obtained in the product of Furukawa et al. The total number of cells in one macromass construct of the present invention is at the least about 132000 which can be much higher also.

Additonally, the physical forms of the products of the Furukawa paper and the present invention are very different; the macromass construct is sheet-like immediately upon formation, whereas the Furukawa aggregates are not sheet-like as is evident from the figures of the respective products.

Further the product of Furukawa differs in diameters with each experiment, as stated in their paper, depending on the confluence of cells used in preparing aggregates whereas the diameter of the construct obtained in the present application does not differ with the confluence of the cells.

The distinct features of the product of the present invention as compared with Furukawa is highlighted in the table below:

Furukawa reference	**Our interpretation**	**Distinct features of the product of the present invention**
Three dimensional fibroblasts aggregates.	The product of the Furukawa et al method is numerous small cell aggregates, which are not unified; they are separate from each other. All cells seeded in the culture vessel do not become part of one aggregate, but are distributed among numerous aggregates.	The product of macromass culture is a single, unified sheet-like construct, entire number of seeded cells in the culture vessel become a part of which. Thus all the seeded cells are integrated into only one unified macroscopic construct. Thus, the Furukawa et al aggregates are not the same as the tissue constructs of the present invention.
Size of one aggregate is 0.2-0.28mm	The size is at the least 10 times less than the lower limit of the macromass construct of the present invention	Size of one construct is at the least greater than 3-5mm in diameter and can be made as large as 2.0 cms as illustrated in our figures, which is 100 times larger than the Furukawa et al aggregates.
Total number of cells in one aggregate of 240 um is about 4000 cells	The number of cells is much lesser than the cells in the macromass tissue construct	The number of cells in one macromass construct is at least 132000 in the smallest macromass construct, and is 9.6×10^6 in a 2.0 cm macromass construct.
The aggregates are not sheet-like in nature.	The physical form of the Furukawa aggregates is markedly different from that of the product of the present invention.	The macromass construct is sheet-like immediately upon formation.
The diameter of the aggregate differs with the confluence of the cells	The diameter is variable with the confluence of the cells	The diameter does not differ with the confluence of the cells.

Additionally, the process as described in the Furukawa et al. reference is also different.

The process of Furukawa has highlighted the following main features of their process in their title /abstract:

1. Rotational culture
2. declining cell- material interaction by using non adhesive substratum
3. insulin, dexamethasone , fibroblast growth factor

As is known that the title and abstract of any paper projects the key findings of the work.

These above features are highlighted in the title and abstract of the Furukawa's cited reference. Hence any reader of this article gets an impression that these factors are paramount in the formation of the aggregates, since these factors are emphasized in the title/abstract, in which high density is not emphasized.

Therefore it cannot be anticipated from the Furukawa et al. paper that tissue like organization of cells can be formed even if all or any of the factors 1, 2, 3 are not used. There is no hint in the paper that high density *alone* can achieve tissue like formation.

The present invention does not employ any of the above features like rotational culture, non adherent substratum, insulin, dexamethasone and basic fibroblast growth factors.

The present invention employs only high-density cell seeding for the formation of the three-dimensional constructs. Thus it requires a unique and distinctly different approach or thinking which is not even mentioned or hinted or anticipated by the cited reference of Furukawa.

Further, it was the main aim of the present invention to form a single unified sheet which includes all the cells which are seeded unlike the numerous separated aggregates of the Furukawa et al. reference. Although Furukawa et al has used a similar high density, there is no clue in their paper that keeping a high density culture static would result in the

end result of a single unified construct. Thus it is the inventive thinking of the inventor of the present invention to perform static high seeding density culture, i.e. macromass culture with the intention and endeavor to form single unified constructs by this different approach for harnessing cell-cell interactions. By following the method of Furukawa et al, it is not possible to produce a single, unified construct, in which all cells seeded in the culture vessel have become integrated into. The Furukawa et al paper does not teach the idea of keeping a high density culture static, on the contrary, it stresses on movement (by rotation). Thus, it cannot be anticipated from their paper, that by keeping a high density culture static, a single, unified construct will form; which is thus the separate inventive thinking of the present inventor. Single, unified 3D constructs which include all seeded cells have hitherto been made by using scaffolds, but in the present invention this has been achieved without the use of any external agent or influence including scaffolds or matrices.

Summary

The applicant hereby summarizes the response by highlighting herein the uniqueness of the product of this invention. The product is a single, unified construct which is three dimensional in nature made of dermal fibroblasts and does not require any external agent or influence that aids in formation of three-dimensional organization of cells.

Further the product is prepared solely by using high density cell seeding wherein the cells are seeded at a high density per unit area of a culture vessel in a range from about $3x10^5$ cells per cm^2 to about $3x\ 10^6$ cells per cm^2, and all the cells seeded in the culture vessel well become a part of the single, unified construct.

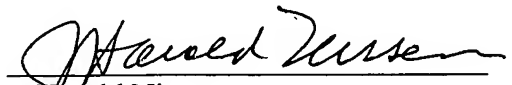
It is respectfully submitted that the above- identified application is now in a condition for allowance and favorable reconsideration and prompt allowance of these claims are respectfully requested.

If the examiner believes that further submissions are required in order to place the application in better condition for allowance, the Examiner is invited to contact the Applicant's undersigned attorney.

If any fees are needed, please charge them to our Deposit Account 50-3108.

Respectfully submitted,

LACKENBACH SIEGEL, LLP
Attorneys for Applicant(s)

By: 

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